Landscape and Watershed Processes

Earthworm Additions Affect Leachate Production and Nitrogen Losses in Typical Midwestern Agroecosystems

William D. Shuster,* Martin J. Shipitalo, Scott Subler, Susanne Aref, and Edward L. McCov

ABSTRACT

Earthworms affect soil structure and the movement of agrochemicals. Yet, there have been few field-scale studies that quantify the effect of earthworms on dissolved nitrogen fluxes in agroecosystems. We investigated the influence of semi-annual earthworm additions on leachate production and quality in different row crop agroecosystems. Chisel-till corn (Zea mays L.)-soybean [Glycine max (L.) Merr.] rotation (CT) and ridge-till corn-soybean-wheat (Triticum aestivum L.) rotation (RT) plots were arranged in a complete randomized block design (n = 3) with earthworm treatments (addition and ambient) as subplots where zero-tension lysimeters were placed 45 cm below ground. We assessed earthworm populations semi-annually and collected leachate biweekly over a three-year period and determined leachate volume and concentrations of total inorganic nitrogen (TIN) and dissolved organic nitrogen (DON). Abundance of deep-burrowing earthworms was increased in addition treatments over ambient and for both agroecosystems. Leachate loss was similar among agroecosystems, but earthworm additions increased leachate production in the range of 4.5 to 45.2% above ambient in CT cropping. Although leachate TIN and DON concentrations were generally similar between agroecosystems or earthworm treatments, transport of TIN was significantly increased in addition treatments over ambient in CT cropping due to increased leachate volume. Losses of total nitrogen in leachate loadings were up to approximately 10% of agroecosystem N inputs. The coincidence of (i) soluble N production and availability and (ii) preferential leaching pathways formed by deep-burrowing earthworms thereby increased N losses from the CT agroecosystem at the 45-cm depth. Processing of N compounds and transport in soil water from RT cropping were more affected by management phase and largely independent of earthworm activity.

EARTHWORMS are well known for their ability to integrate the physical, chemical, and biological domains of the soil ecosystem (Lee, 1985; Edwards and Bohlen, 1996). Moreover, earthworms are present in many agroecosystems where they influence nutrient cycling and hydrologic processes (Shipitalo and Edwards, 1993; Li and Ghodrati 1995; Subler et al., 1997). Consequently, earthworms influence the retention or loss of dissolved forms of nutrients, with implications for nutrient management in agroecosystems. Yet, the relation-

W.D. Shuster, S. Subler, and E.L. McCoy, School of Natural Resources, The Ohio State University, OARDC, Wooster, OH 44691. M.J. Shipitalo, USDA Agricultural Research Service, North Appalachian Experimental Watershed, Coshocton, OH 43812. S. Aref, Department of Statistics, University of Illinois, Urbana, IL 61820. Received 8 Aug. 2002. *Corresponding author (shuster.william@ena.gov).

Published in J. Environ. Qual. 32:2132–2139 (2003). © ASA, CSSA, SSSA 677 S. Segoe Rd., Madison, WI 53711 USA ship between earthworm abundance, species distribution, and their habitat in agroecosystems is interactive. The amount, type, and quality of residues left on the soil surface; soil and climate conditions; the type and frequency of tillage; and the type and extent of fertilizer and pesticide application all play a role in the regulation of both earthworm activity and their community structure in agroecosystems.

Tillage affects both earthworm population dynamics and patterns in solute transport. Several workers have shown that no-till management increases the abundance of earthworms and can also enhance drainage characteristics (Edwards et al., 1989, 1992; Bicki and Guo, 1991; Trojan and Linden, 1998). On the other hand, periodic disturbances in the form of chisel-tillage will temporarily aerate soils but also destroy pores formed by roots and earthworm burrows. Earthworms also have more direct effects on soil macroporosity. The semi-permanent burrows created by *Lumbricus terrestris* have profound effects on solute transport by providing preferential flow pathways that can connect surface soils with shallow water tables (Shipitalo and Edwards, 1996; Li and Ghodrati, 1995; Lachnicht et al., 1997; Subler et al., 1997).

The activities of deep-burrowing (anecic) earthworms concomitantly create preferential pathways for water and solutes, where they also influence nitrogen cycling in the soil ecosystem (Subler et al., 1997, 1998; Helling and Larink, 1998; Hendrix et al., 1998; Whalen et al., 1999). The preferential flow paths formed by deep-burrowing earthworms and other macroinvertebrates have also been found to be biological "hot spots" in soils, with correspondingly higher levels of microbial activity (Parkin and Berry, 1999; Bundt et al., 2001). The earthworm species L. terrestris overlay their deep burrows with structures known as middens, which are accumulations of raw and decomposed crop residues, casts, and soils (Nielsen and Hole, 1964). This arrangement brings organic matter, earthworms, and the microbial biomass into intimate contact, accelerating decomposition and therefore affecting nutrient cycling processes (Subler and Kirsch, 1998).

Nitrogen compounds are subject to dissolution and transport out of the agroecosystem via runoff and denitrification, or when they are leached below the crop rooting zone. Leachate contaminated with N compounds is a degrading influence on surface waters and ground water reserves (Ward et al., 1994; Owens et al.,

Abbreviations: CT, chisel till, corn–soybean rotation; DON, dissolved organic nitrogen; RT, ridge till, corn–soybean–wheat rotation; TIN, total inorganic nitrogen.

1995; Wilkison and Blevins, 1999). The negative effects of nitrogen contamination on both human health and the environment at-large are presently the subject of much controversy. Thus, it is of practical interest whether the extent of leaching losses of dissolved nutrients from agricultural systems can be influenced by either earthworm communities, the agroecosystems that they inhabit, management phase, or interactions among these factors. Lysimetry provides a means to assess total fluxes of water and dissolved nitrogen compounds through the soil matrix (Owens et al., 1995). Therefore, we studied the effects of both ambient and manipulated earthworm communities, which were altered through the addition of deep-burrowing earthworms, on typical Midwestern production agroecosystems, and sampled over the long term to determine the potential for these treatments to affect the volume and quality of leachate collected in zero-tension pan lysimeters.

MATERIALS AND METHODS

Site Description

We conducted the study from October 1994 to December 1997 at the Ohio Management Systems Evaluation Area (MSEA) (Ward et al., 1994) near Piketon, OH (39°02' N, 83°02′ W). Soils at this flood plain site are predominantly Huntington silt loam (fine-silty, mixed, active, mesic Fluventic Hapludoll), with landscape slopes ranging between 2 to 5%. These are shallow soils with a maximum A horizon depth of approximately 60 cm and an abrupt transition to a gravelly B horizon. A typical particle-size analysis of the upper 15 cm of soil was 210 g kg⁻¹ sand, 550 g kg⁻¹ silt, and 240 g kg⁻¹ clay (Nokes et al., 1997). Soil organic carbon concentrations average 15.8 g kg⁻¹ in the surface 15 cm. Average annual precipitation at this site is 900 mm, with 570 mm of this falling between May and November. The water table is rarely more than 2 m below ground surface. On average, there are 170 d yr⁻¹ without a killing frost (Nokes et al., 1997).

Cropping system treatments were established in 1991 and were a corn-soybean rotation (CT) and a corn-soybeanwinter wheat-hairy vetch (Vicia villosa L.) cover-crop rotation (RT). The CT corn crop was planted with a no-till planter into soybean stubble. In preparation for the soybean crop, soil was chisel-disked (25-cm depth, one pass), then offsetdisked (10-cm depth, two passes). The RT system was based on permanent ridges that were built in October 1990 to a height of 20 cm with a ridge-till planter. Corn and soybean crops were cultivated twice in RT; ridges were rebuilt on the second cultivation. Wheat was drilled on and between the ridges remaining from the soybean crop. Vetch was planted after wheat harvest in July, then killed with glyphosate [N-(phosphonomethyl) glycine] before planting corn the next season. Details on nitrogen inputs are shown in Table 1. Each management system was replicated (n = 3) in 0.4-ha plots, which were arranged in a complete randomized block design, with a split plot for earthworm treatment.

In each of the three replicate cropping system blocks, one pair of ambient and addition enclosures was designated for earthworm population assessments, while another pair was instrumented with lysimeters.

Earthworm community manipulations were ambient and addition. The ambient population was used as a control in this experiment; this treatment received no additions of earthworms. Earthworm additions were made to enclosures that measured 6.1 m on a side and consisted of corrugated plastic sheeting extending 25 cm above the soil surface and to a depth of approximately 5 cm. Each spring and fall, earthworms were added at a rate of 100 individuals m⁻². Earthworm additions were initiated in the fall of 1993 and were continued through the fall of 1997. Earthworms used for addition treatments were collected by formalin expulsion (Raw, 1962) from a notill corn field in Columbus, OH. Earthworm addition groups contained an average of 76% adult and juvenile *L. terrestris* individuals; the remaining earthworms were *Aporrectodea* or *Octolasion* spp. (Subler et al., 1997).

Earthworm populations were assessed before semi-annual additions and in the spring and fall of each year from fall 1994 to spring 1997. We sampled for earthworms by removing soil from four pits (38 by 38 by 15 cm deep). This soil was handsorted for earthworms. Deep-burrowing earthworms were driven out of their burrows by saturating the excavated soil surface at a 15-cm depth with dilute formalin (Raw, 1962). All earthworms that surfaced within 30 min were collected. All earthworms were preserved in 5% formalin, and were identified to the level of species (adults) or genus (juveniles) (Schwert, 1990).

Zero-tension pan lysimeters (38 cm square and 4.5 cm deep) were constructed from high-density polyethylene (HDPE). The lysimeters were filled just below their top edge with acid-washed quartz gravel and plumbed with HDPE tubing to drain into a 20-L glass carboy. Lysimeters were installed nondestructively 45 cm below ground surface in each earthworm treatment plot after planting in June 1994. The pans were aligned so that one edge was directly under and parallel to a crop row and the opposite edge was directly under the adjacent midrow position. To complete lysimeter installation, access tubes were extended from the glass collection jar to the soil surface. Leachate was collected with a vacuum sampling device into acid-washed glass containers.

Lysimeters were sampled twice per month between October 1994 and December 1997. Additional samplings were made after major storms. Leachate volume was measured and subsamples were analyzed for inorganic and organic forms of nitrogen. Leachate total inorganic nitrogen (TIN) concentrations were the sum of NH₄–N, NO₂, and NO₃ concentrations. Nitrate, nitrite, and ammonium concentrations were determined with phenate and cadmium reduction–diazotization methods on a flow-injection analyzer. Dissolved organic nitrogen (DON) was calculated as the difference between the TIN concentration and the NO₃–N concentration determined after

Table 1. Management schedule for cropping systems at the Management Systems Evaluation Area (MSEA), Piketon, Ohio, 1994 to 1997.

Agroecosystem	1994	1995	1996	1997	
Chisel tillage (CT)					
Crop	corn	soybean	corn	soybean	
N inputs, kg N ha ⁻¹	30†; 120 sidedress†	· –	30; 120 sidedress	-	
Ridge tillage (RT)					
Crop	corn	sovbean	wheat-vetch	corn	
N inputs, kg N ha ⁻¹	30†; 28 (vetch)‡; 90† sidedress	_	_	30; 28 (vetch); 90 sidedress	

[†] Nitrogen applied as 30% urea solution.

Average vetch N contribution according to W. Lewis (personal communication).

alkaline persulfate digestion of leachate samples (Cabrera and Beare, 1993). Biweekly data were pooled by management-hydrologic quarters: late growing (LG), August to October; early dormant (ED), November to January; late dormant (LD), February to April; and early growing (EG), May to July. In this study, each quarter is followed by the numerals 1, 2, 3, and 4 to designate the time intervals 1994 to 1995, 1995 to 1996, 1996 to 1997, and 1997 to 1998, respectively. Quarterly mean concentration and total load for TIN and DON were calculated for each treatment.

Earthworm abundance, leachate volume, and flow-weighted TIN and DON data were \log_{10} -transformed to satisfy assumptions of analysis of variance (ANOVA). Treatment effects were tested for significance with mixed-model analysis of variance (PROC MIXED; SAS Institute, 2002). For variables with significant cropping system effects, separate models were run to analyze interactions between management phase and earthworm treatment by cropping system. The threshold for significance was set at $P \le 0.05$ unless indicated otherwise.

RESULTS AND DISCUSSION Earthworm Community Manipulations

Semi-annual earthworm population assessments indicated that the abundance of L. terrestris was increased from either zero or very low levels with earthworm additions, and for both agroecosystems (Table 2). Populations of L. terrestris were generally higher in RT treatments compared with CT (Table 2), yet this trend was significant for only the first two assessments. Although the populations of deep-burrowing species were generally small relative to the numbers added, the spring assessments sampled more earthworms overall than those made in the fall. For the spring samplings, an increased abundance of deep-burrowing species was apparently at the expense of surface-dwelling species (i.e., epigeic) like L. rubellus. Although the decline in epigeic populations was significant for the spring 1996 assessment only, this trend is consistent with the short-term assessments of Subler et al. (1997) whereby additions of deep-burrowing earthworms apparently decreased surface-dweller populations, and to a greater extent in CT than RT cropping. We speculate that the decline in epigeic earthworms was probably due to removal of crop residues from the soil surface by *L. terrestris*. The abundance of soil-dwelling endogeic species was zero in the first few assessments, then increased overall during the course of the study and with somewhat greater mean numbers of individuals sampled in RT than CT cropping (Table 2).

Semi-annual earthworm population assessments indicated that *L. terrestris* populations were highly variable (Table 2). The deep-burrowing species that we did sample accounted for only a small proportion of the added earthworms. Although populations were generally and significantly increased in addition over ambient treatments, it is questionable whether these populations reached steady state at any point in this experiment. Other workers have found that various methods used to introduce non-native earthworm species to agroecosystems or amend existing populations failed to establish even relatively small populations (Butt et al., 1999).

Since our earthworm treatments involved forcing a large added population on both an existing earthworm community and a relatively fixed resource base, all populations were probably poorly accommodated for some period of time. These conditions may have led to increased burrowing and foraging shortly after additions were made. Added earthworms may also have dispersed outside of the plots, or burrowed deep enough to avoid sampling. The efforts of individuals that were temporarily active may have increased both the abundance of semi-permanent macropores and short-term residue consumption. It is also possible that our large additions triggered competition for scarce resources among and between populations of deep-burrowers and surface dwellers. Our results would probably have been different if earthworm populations had been allowed to in-

Table 2. Mean earthworm abundance for the predominant species of surface-dwelling (epigeic), deep-burrowing (anecic), and soil-dwelling (endogeic) earthworms in the ambient and addition treatments for the two agroecosystems.

Agroecosystem†	Earthworm treatment	Addition groups							
		Fall 1994	Spring 1995	Fall 1995	Spring 1996	Fall 1996	Spring 1997		
			——————————————————————————————————————						
			Lumbricus rubellus, epigeic						
CT	ambient	0a,x‡	12.2a,x	1.2a,x	60.1a,x	0.6a,x	19.1a,x		
	addition	0.6a,x	1.2a,x	3.9a,x	6.5b,x	1.2a,x	7.8a,x		
RT	ambient	3.3a,x	46.1a,x	0.6a,x	49.1a,y	4.5a,x	40.2a,x		
	addition	3.6a,x	15.2a,x	0a,x	36.0b,y	0.6a,x	15.5a,x		
		L. terrestris, anecic							
CT	ambient	0a,x	0a,x	0a,x	2.4a,x	0a,x	0a,x		
	addition	0.6b,x	3.6b.x	7.8b,x	6.7a,x	0a,x	3.0b,x		
RT	ambient	0.6a,y	0a,y	0a,x	1.8a,x	0a,x	0a,x		
	addition	7.1b,y	17.9b,y	6.0b,x	5.7a,x	1.8a,x	3.0b,x		
		Octolasion tyrtaeum, endogeic							
CT	ambient	0a,x	0a,x	0a,x	0a,x	5.4a,x	6.5a,x		
	addition	0a,x	0a,x	0a.x	1.8a,x	8.7a,x	6.0a,x		
RT	ambient	0a,x	0a,x	0a.x	0a,x	3.6a,x	29.2a,x		
	addition	0a,x	0a,x	0.6a,x	1.8a,x	17.6a,x	9.8a,x		

 $[\]dagger$ CT, chisel till, corn–soybean rotation; RT, ridge till, corn–soybean–wheat rotation.

[‡] Means in the same column followed by different letters (a and b) within species and addition treatments are significantly different at the 0.05 probability level. Means in the same column followed by different letters (x and y) within species and agroecosystems are significantly different at the 0.05 probability level.

crease under more natural conditions and in response to an increased availability of food resources. Our results also show, however, that a relatively large number of earthworms were probably not necessary to maintain the small populations of anecic species that typically coexist with species from other ecological groups, which comprise the larger proportion of individuals at this site. In addition, our observations of earthworm community dynamics may be an indication that populations of deepburrowing earthworms were not well-adapted to aspects of the flood plain environment. Above and beyond the significant influence of climate and different tillage regimes, other effects on establishment would include shallow water tables and soil depths of approximately 50 cm, which may limit the depth of burrowing; an increased frequency of flooding; higher predation due to increased activity at the surface of a saturated soil; or stresses related to sampling.

Leachate Production

Leachate production in both systems was greatest when soils were near saturation in the early spring and winter. Increased evapotranspiration during the growing seasons typically led to a decrease in leachate production. The amount of leachate produced in CT and RT treatments was similar in magnitude (Table 3; P = 0.73). Yet, leachate production was significantly affected by the addition of deep-burrowing earthworms in CT cropping only (P < 0.0001). When normalized for total precipitation received in a given quarter, we found that the addition of deep-burrowing earthworms significantly increased the amount of leachate lost in CT cropping 3-fold for the smallest quarterly rainfall (i.e., LD1) and 30-fold during the quarter with highest precipitation (i.e., EG2). These ranges were much narrower in RT,

where the proportions of leachate lost were similar in ambient and addition treatments for the highest quarterly rainfall. Agroecosystems also responded differently to stress of major rainfalls. For example, an 85-mm rainfall with an approximate 24-h duration in EG3 registered as a 150-fold increase in leachate lost from CT treatments where earthworms were added than ambient, whereas in RT cropping, earthworm treatments produced nearly equal proportions of leachate.

The contrasts between earthworm treatments in CT cropping are ascribed to preferential flow regimes in addition treatments. Macroporosity was probably increased by the significantly higher populations of deepburrowing L. terrestris. The network of semi-permanent burrows would serve to drain ponded areas and variably saturated soils (McCoy et al., 1994) through bypass flow. Leachate production was also a function of management phase, which integrates the effects of both season (i.e., hydrologic quarter) and crop-tillage combinations specific to a particular rotation. For example, annual tillage in CT may have temporarily increased structural macroporosity and decreased earthworm populations through mechanical dismemberment. Structural macroporosity, however, may have been balanced by the concomitant destruction of the proportion of previously established earthworm burrow systems closer to the soil surface. The nature of our earthworm addition treatments may also have produced large numbers of individuals that were potentially burrowing and foraging more than they would under more amenable resource regimes. Limits on resource quality and availability in CT may have spurred these transient populations to burrow more frequently within the treatment plots, at least until they expired.

For treatments without added earthworms, leachate

Table 3. Precipitation and leachate production as a proportion of quarterly precipitation for each agroecosystem and earthworm treatment.

		Chis	el-till	Ridg	ge-till
Management phase†	Total precipitation	Ambient	Addition	Ambient	Addition
	mm			% —	
ED1	256	4.3	15.1**	8.6	12.5
LD1	172	4.6	15.8**	5.2	11.8
EG1	302	1.6‡	10.4**	4.4‡	4.7
LG1	357	1.4 ‡	4.5**	3.9‡	4.7
ED2	224	1.8	12.2**	10.5	7.2
LD2	232	1.1	19.8§	13.8	20.7
EG2	404	0.3¶	9.3*	5.0¶	4.4
LG2	176	1.0¶	6.8§	7.7¶	3.4
ED3	243	1.4	11.4*	10.2	7.1
LD3	279	0.5	8.1*	8.8	18.9
EG3	373	0.3‡	45.2§	9.9#	10.2
LG3	211	10.2‡	5.1**	2.1#	3.2
ED4	218	2.2	27.6**	10.7	5.9
Mean	265	2.4	14.7	7.8	8.8
Minimum	172	0.3	4.5	2.1	3.2
Maximum	404	10.2	45.2	13.8	20.7

^{*} Significant at the 0.05 probability level.

^{**} Significant at the 0.01 probability level. *** Significant at the 0.001 probability level.

[†] ED, early dormant period; LD, late dormant period; EG, early growing period; LG, late growing period. The numerals 1, 2, 3, and 4 to designate the time intervals 1994 to 1995, 1995 to 1996, 1996 to 1997, and 1997 to 1998, respectively.

[§] Significant at the 0.10 probability level.

[¶] Corn.

[#] Wheat.

Table 4. Time-averaged (1994–1997) leachate concentrations (n = 3) of total inorganic nitrogen (TIN) and dissolved organic nitrogen (DON) for agroecosystems (CT, chisel till, corn-soybean rotation; RT, ridge till, corn-soybean-wheat rotation) and earthworm treatments.

Management phase†	TIN				DON				
	CT		RT		CT		RT		
	Ambient	Addition	Ambient	Addition	Ambient	Addition	Ambient	Addition	
	mg L ⁻¹								
ED1	0.5	2.7	5.4	2.8	0.1	1.6	3.1	0.7	
LD1	3.3	3.2	2.9	1.4	5.5	3.6	1.9	0.7	
EG1	2.2‡	2.3	3.2 ‡	1.6	0.9‡	3.0	1.4‡	1.0	
LG1	3.0‡	8.1	2.8‡	5.0	2.4‡	2.8	3.1‡	7.0	
ED2	4.1	12.7	6.0	8.0	2.7	5.8	3.3	2.6	
LD2	1.2	9.5	6.8	9.9	0.6	2.8	2.8	4.1	
EG2	1.0§	7.4	3.8¶	2.6	<0.1§	1.1	0.5¶	0.4	
LG2	1.0§	5 . 5	8.2¶	9.6	0.5§	1.0	1.8¶	3.0	
ED3	7.1	12.2	3.2	13.1	1.4	5.4	1.0	11.1	
LD3	5.8	8.8	2.3	9.6	1.6	4.4	1.3	6.2	
EG3	2.2‡	4.7	9.0§	7.9	2.0‡	3.1	5.3§	4.8	
LG3	1.8‡	2.4	1.1§	3.1	0.7 ‡	3.1	1.9§	1.2	
ED4	4.1	8.8	5.4	3.8	2.2	4.2	5.8	3.3	
Mean	2.9	6.8	4.6	6.0	1.6	3.2	2.6	3.5	
Minimum	0.5	2.3	1.1	1.4	< 0.1	1.0	0.5	0.4	
Maximum	7.1	12.7	9.0	13.1	5.5	5.8	5.8	11.1	

[†] ED, early dormant period; LD, late dormant period; EG, early growing period; LG, late growing period. The numerals 1, 2, 3, and 4 to designate the time intervals 1994 to 1995, 1995 to 1996, 1996 to 1997, and 1997 to 1998, respectively.

production tended to be higher in RT than CT. This suggests that earthworm ambient plots in RT conducted, qualitatively, more water than corresponding plots in CT cropping (Table 3). This is consistent with the results of double-ring infiltration measurements (Shuster, 2000) at the site, which showed that cumulative infiltration capacity averaged 25% greater in RT than in CT cropping. We note here that management phase was an overall more significant influence on leachate production in RT (P = 0.04) than CT cropping (P = 0.07). Other workers have also reported that RT cropping generally promotes infiltration (Clay et al., 1992; Kanwar et al., 1985) through a reduction or elimination of disturbance from tillage, freer conduction through senescent root zones, enhanced activity across all earthworm ecological groups, and cracking or ripening of the soil structure. The soil structure thus developed in RT may have either augmented or deemphasized the effects of any additional macropores in the addition treatments. A consequence of these conditions in RT earthworm treatments was a trend toward elevated populations for both surface- and soil-dwelling species (Table 2). Earthworm food resources in RT cropping were of greater variety, more abundant, and probably more evenly distributed. The increased availability of food resources in RT than CT cropping may have also contributed to a more random distribution of burrows in RT. Cook and Linden (1996) found that epigeic and endogeic species create numerous burrow networks in the surface 20 cm of soil, yet burrowing habits became considerably less random when food resources are encountered. It stands to reason that a more even distribution of different diameters of burrows was produced in RT than CT cropping, influencing movement of water in RT cropping under both unsaturated and saturated conditions.

Leachate Quality and Loadings

Mean concentration of TIN ranged between 0.5 to 13.1 mg L^{-1} and differed on the basis of management phase (P=0.05) and earthworm treatment in CT cropping only (P=0.02). Mean concentration of dissolved organic nitrogen ranged from <0.01 to 11 mg L^{-1} and was only weakly affected by management phase in the CT rotation (P=0.08). Earthworm addition treatments in RT cropping had nearly double the maximum concentration of DON than CT, though this difference was not significant (P=0.64). Both TIN and DON were concentrated in the second and third early dormant (ED) management phases for both agroecosystems, and especially where earthworms were added (Table 4).

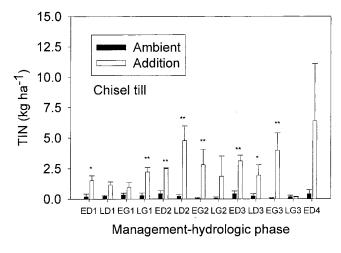
Loadings of dissolved nitrogen compounds in leachate from CT and RT cropping were similar (Fig. 1 and 2; P = 0.64). Losses of TIN on the basis of management phase were not strongly significant for CT (P =0.09), although they were for RT cropping (P < 0.05). For CT cropping, earthworm additions altered soil water balance over the greater proportion of management phases and evidently in a manner that favored preferential flow pathways, which led to higher loadings of dissolved TIN (Fig. 1; P = <0.0001). Leachate loadings of TIN (Fig. 1; P = 0.75) and DON (Fig. 2; P = 0.70) for RT cropping were generally independent of earthworm treatments. Management phase, however, had a weak effect on DON losses for CT cropping only (P = 0.08). Yet, differences in TIN and DON losses from earthworm treatments in RT cropping were significant only for flood conditions encountered during LD3, wherein 152 mm rain fell over a 2-d period.

Soluble nitrogen compounds were produced to different extents though at roughly similar times in CT and RT cropping and were apparently regulated at the

[‡] Soybean.

[§] Corn.

[¶] Wheat.



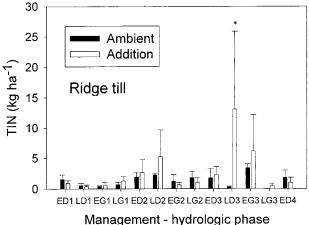
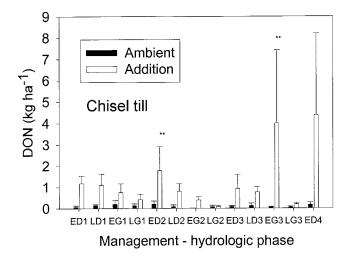


Fig. 1. Effects of earthworm treatments on losses of total inorganic nitrogen (TIN) in leachate from cropping systems at the Piketon, Ohio site. Error bars indicate one standard error. The terms *, ***, and *** indicate significant differences at the 0.05, 0.01, and 0.001, probability levels, respectively.

agroecosystem level. Although dissolved nitrogen concentrations in leachate varied widely, their cumulative loadings revealed significant losses from agroecosystems. The leaching losses observed in this study were approximately 7 kg N ha⁻¹ yr⁻¹, which accounts for a relatively small proportion of annual N inputs to these production agroecosystems, where inputs averaged 150 kg N ha⁻¹ yr⁻¹ for the corn crops at our Midwestern U.S. study site (Table 1). Our observed nutrient losses in leachate were similar, however, to those observed by de Vos (2000), who reported nitrate losses on the order of 10 kg N ha⁻¹ yr⁻¹ for tiles draining a silt loam soil.

Management phase and its correspondent hydrologic regime in each agroecosystem regulated the extent and significance of earthworm effects, and particularly in CT. It appears that the different phases of each cropping system have major influences on TIN losses. For example, CT cropping was charged with concentrations of TIN in the early dormant phases for each year of this experiment. Crop senescence is a likely explanation for this, as this period involves relatively little nutrient uptake and a sharp decline in plant water use. The majority of soluble nitrogen compounds would then be more



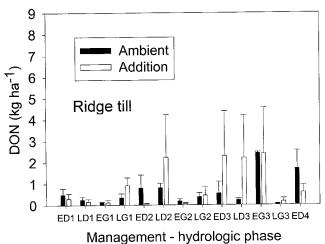


Fig. 2. Effects of earthworm treatments on losses of dissolved organic nitrogen (DON) in leachate from cropping systems at the Piketon, Ohio site. Error bars indicate one standard error. The terms *, **, and *** indicate significant differences at the 0.05, 0.01, and 0.001, probability levels, respectively.

available for transport than uptake, as expected during the growing season. Losses were relatively high for specific quarters (e.g., LD2, LD3, EG3, and ED4). This is similar to patterns observed in leachate production, where management phase and earthworm additions interacted; this effect extended to CT cropping only. The late dormant and early growing phases correspond to periods of higher earthworm activity, low disturbance, and higher levels of nitrate availability. Precipitation added to already saturated soils in the late dormant phase acts to flush nitrates accumulated after crop senescence (Owens et al., 1995). This may have been particularly true for CT cropping where both tillage and earthworms (Hendrix et al., 1998) can move N compounds deeper into the soil profile. During the early growing season, when deep-burrowing earthworms become active, the inorganic N fertilizers added at planting or as sidedress can increase the availability of mineral N (Helling and Larink, 1998; Subler et al., 1998). This N is available before the growing crop has developed an ability to uptake the nutrients. Soybean cropping exhibited slightly less TIN than corn cropping phases in both CT and RT; the absence of N inputs and basic differences in the way that soybeans cycle N are likely explanations for this. In contrast to TIN production and losses, management phase had a significant effect on the loss of DON in RT only. Conditions in RT cropping may create more opportunities for both water and nitrogen to be scavenged by successive crops and would also alter the extent to which water and nitrogenous solutes were available for transport.

Earthworm addition treatments in CT cropping consistently increased the N loading in leachate. The concentration of TIN in leachate may have been increased by transport of fertilizer-derived N or by greater mineralization of organic matter in earthworm burrows. Earthworm burrows and soils enriched with casts are known to provide habitat for nitrifying bacteria and are enriched with higher levels of nitrate than the surrounding bulk soil (Blair et al., 1995; Parkin and Berry, 1999). Yet, the influence of earthworm additions on flow-weighted TIN losses was more variable in RT (Fig. 2) than CT cropping. The combination of different flow regimes from a ripened soil structure and production processes in RT versus CT cropping probably increased variability in losses of nitrogen compounds from the ridge-till agroecosystem. The DON losses from RT may reflect the role that earthworms can play in the decomposition of a more or less continual supply of organic matter resources (Subler and Kirsch, 1998). Under higher soil water contents in RT cropping, this would serve to provide suitable habitat and substrate for microbes, thereby increasing their activity. Although Parkin and Berry (1999) found that earthworm burrows were enriched in nitrate and nitrifying bacteria, they did not measure burrow soil for organic N compounds. The organic N present in earthworm burrows may have been available for nitrification, thereby decreasing the proportional amount of DON in leachate and increasing the amount of nitrate. In combination with variance over our long-term assessments, proportional shifts among N pools may explain the contrasting results of Subler et al. (1997), which registered higher DON losses (i.e., compared with TIN) for one week of leachate collection during the late growing season in 1994. These observations in RT contrast with those in CT cropping where shifts in earthworm community structure evidently changed the nature of flow regimes.

Nitrogen compounds made available from semiannual earthworm additions may have confounded observations of TIN and DON concentrations in agroecosystems (Table 4). Blair et al. (1995) pointed out that earthworms contribute high secondary productivity to nitrogen cycles in agroecosystems with upper-bound estimates of turnover approaching 60 kg total N ha⁻¹ yr⁻¹. Major proportions of this turnover result from the nearcontinual production of labile N from sources that include earthworm mucous exudate, casts, urine, and dead earthworm tissue. Assuming that all earthworms died immediately after they were added to the agroecosystems, our earthworm additions would have added the equivalent of 50 kg TIN ha⁻¹ yr⁻¹. This figure indicates a nearly fourfold increase in nitrogen over that observed for annual total N losses in leachate. Although many earthworms expired, we know that this was not case for all individuals. Some unknown proportion of these individuals may have also escaped from the plots or were preyed on. We also note that losses of leachate and dissolved nitrogen compounds were similar between agroecosystems, which had different relative abundances of earthworm species. Although this N would have been available a few weeks after additions were made, a proportionate magnitude of loss is not reflected in time-averaged leachate concentrations for TIN or DON, which themselves did not differ on the basis of earthworm treatments. In a microcosm study, Whalen et al. (1999) showed that most N derived from earthworm tissue was rapidly turned over and these predominantly organic forms were then taken up by plant shoot biomass. These results predict that little N would be available for dissolution and leaching. Nitrogen that was not leached would have been stored in soil N pools, sequestered in crop or microbial biomass, or lost through denitrification pathways during the wetter dormant phases. We concluded that there was no apparent artifact from earthworm additions contributing to variance in TIN or DON loads from either agroecosystem studied.

CONCLUSIONS

These results indicate that contrasting cropping conditions and alterations in earthworm community structure affected both leachate production and quality in typical Midwestern U.S. tillage systems. Additions of deep-burrowing earthworms led to overall leachate losses at a 45-cm soil depth, which ranged overall between 3.2 and 45.2% of measured precipitation. Dissolved nitrogen loadings were found to overall account for up to approximately 7% of total agroecosystem N inputs. Shifts in earthworm community structure with additions of deep-burrowing earthworms apparently patterned leachate production and nutrient cycling in a way that corresponded to the ecological habits of dominant earthworm species.

Periodic disturbance in the CT system highlights the effects of adapted species, such as *L. terrestris*, and their deep-burrowing habit and relatively aggressive foraging.

Our approach to amending earthworm populations may have created a transient population of anecic individuals, which could have foraged and burrowed intensively. A possible artifact of our earthworm treatments would be a higher degree of macroporosity than ordinarily expected for natural populations supported by a sufficient resource base. For those *L. terrestris* that established, their middens formed "hot spots" of foraged coarse organic matter and N cycling, which overlay preferential flow paths formed by deep-burrowing earthworms. The coincidence of enhanced soluble N production and availability and preferential leaching pathways thereby increased N losses from the CT agroecosystem. Yet, the RT agroecosystem supported a more diverse earthworm community than CT. The inter-

actions of earthworms that occupied either the surface soil, shallow horizons, or both regions probably contributed to more variety in the processing of organic matter and nitrogen compounds. In the case of RT cropping, any additional deep burrows due to earthworm treatments were in addition to the infiltration benefits contributed by a ripened soil structure and more root channels; these factors probably led to an overall greater abundance of tortuous pathways in the surface soil. Higher variance in the processing and transport of soil water and N compounds in RT led to their retention, uptake, or loss to different extents than that observed in CT cropping.

The present study suggests that deep-burrowing earthworms can be a significant influence on leachate production, nitrogen production, and thereby leachate quality. Therefore, the extent of leaching and nutrient loss are regulated by the type of agroecosystem and its productivity, and can be furthermore influenced by its capacity for supporting populations of deep-burrowing earthworms such as *L. terrestris*.

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REFERENCES

- Bicki, T.J., and L. Guo. 1991. Tillage and simulated rainfall intensity effect on bromide movement in an Argiudoll. Soil Sci. Soc. Am. J. 55:794–799.
- Blair, J.M., R.W. Parmelee, and P. Lavelle. 1995. Influences of earthworms on biogeochemistry of North American ecosystems p. 124–158. *In* P.F. Hendrix (ed.) Earthworm ecology and biogeography in north america. Lewis Publ., Ann Arbor, MI.
- Bundt, M., F. Widmer, M. Pesaro, J. Zeyer, and P. Blaser. 2001. Preferential flow paths: Biological 'hot spots' in soils. Soil Biol. Biochem. 33:729–738.
- Butt, K.R., M.J. Shipitalo, P.J. Bohlen, W.M. Edwards, and R.W. Parmelee. 1999. Long-term trends in earthworm populations of cropped experimental watersheds in Ohio, USA. Pedobiologia 43:713-719
- Cabrera, M.L., and M.H. Beare. 1993. Alkaline persulfate oxidation for determining total nitrogen in microbial biomass extracts. Soil Sci. Soc. Am. J. 57:1007–1012.
- Clay, S.A., D.M. Clay, W.C. Koskinen, and G.L. Malzer. 1992. Agrichemical placement impacts on alachlor and nitrate movement through soil in a ridge tillage system. J. Environ. Sci. Health Part B 27:125–138.
- Cook, S.M.F., and D.R. Linden. 1996. Effect of food type and placement on earthworm (*Aporrectodea tuberculata*) burrowing and soil turnover. Biol. Fertil. Soils 21:201–206.
- De Vos, J.A., D. Hesterberg, and P.A.C. Raats. 2000. Nitrate leaching in a tile-drained silt-loam soil. Soil Sci. Soc. Am. J. 64:517–526.
- Edwards, C.A., and P.J. Bohlen. 1996. The biology and ecology of earthworms. Chapman and Hall, New York.
- Edwards, W.M., M.J. Shipitalo, W.A. Dick, and L.B. Owens. 1992. Rainfall intensity affects transport of water and chemicals through macropores in no-till soil. Soil Sci. Soc. Am. J. 56:52–58.
- Edwards, W.M., M.J. Shipitalo, L.B. Owens, and L.D. Norton. 1989. Water and nitrate movement in earthworm burrows within long-term no-till cornfields. J. Soil Water Conserv. 44:240–243.
- Helling, B., and O. Larink. 1998. Contribution of earthworms to nitro-

- gen turnover in agricultural soils treated with different mineral N-fertilizers. Appl. Soil Ecol. 9:323–329.
- Hendrix, P.F., A.C. Peterson, M.H. Beare, and D.C. Coleman. 1998. Long-term effects of earthworms on microbial biomass nitrogen in coarse and fine textured soils. Appl. Soil Ecol. 9:381–386.
- Kanwar, R.S., J.L. Baker, and J.M. Laffen. 1985. Nitrate movement through the soil profile in relation to tillage system and fertilizer application method. Trans. ASAE 28:1802–1807.
- Lachnicht, S.L., R.W. Parmelee, D. McCartney, and M. Allen. 1997. Characteristics of macroporosity in a reduced tillage agroecosystem with manipulated earthworm populations: Implications for infiltration and nutrient transport. Soil Biol. Biochem. 29:493–498.
- Lee, K.E. 1985. Earthworms: Their ecology and relationships with soil and land use. Academic Press, Sydney, Australia.
- Li, Y., and M. Ghodrati. 1995. Transport of nitrate in soils as affected by earthworm activities. J. Environ. Qual. 24:432–438.
- McCoy, E.L., C.W. Boast, R.C. Stehouwer, and E.J. Kladivko. 1994. Macropore hydraulics: Taking a sledgehammer to classical theory. p. 303–348. *In R. Lal and B.A. Stewart* (ed.) Soil processes and water quality. Adv. Soil Sci. Lewis Publ., Ann Arbor, MI.
- Nielsen, G.A., and F.E. Hole. 1964. Earthworms and the development of coprogenous A1 horizons in forest soils of Wisconsin. Soil Sci. Soc. Am. Proc. 28:426–430.
- Nokes, S.E., N.R. Fausey, S. Subler, and J.M. Blair. 1997. Stand, yield, weed biomass, and surface residue cover comparisons between three cropping/tillage systems on a well-drained silt loam soil in Ohio, USA. Soil Tillage Res. 44:95–108.
- Owens, L.B., W.M. Edwards, and M.J. Shipitalo. 1995. Nitrate leaching through lysimeters in a corn–soybean rotation. Soil Sci. Soc. Am. J. 59:902–907.
- Parkin, T.B., and E.C. Berry. 1999. Microbial transformations in earthworm burrows. Soil Biol. Biochem. 31:1765–1771.
- Raw, F. 1962. Studies of earthworm populations in orchards: I. Leaf burial in apple orchards. Ann. Appl. Biol. 50:389–404.
- SAS Institute. 2002. SAS/STAT online user's guide. Ver. 8.02. SAS Inst., Cary, NC.
- Schwert, D.P. 1990. Oligochaeta: Lumbricidae. p. 341–355. *In D.L.* Dindal (ed.) Soil biology guide. John Wiley & Sons, New York.
- Shipitalo, M.J., and W.M. Edwards. 1993. Seasonal patterns of water and chemical movement in tilled and no-till column lysimeters. Soil Sci. Soc. Am. J. 57:218–223.
- Shipitalo, M.J., and W.M. Edwards. 1996. Effects of initial water content on macropore/matrix flow and transport of surface-applied chemicals. Soil Sci. Soc. Am. J. 25:662–668.
- Shuster, W.D. 2000. Influences of earthworm community dynamics on soil structure, carbon distribution, solute transport, and leachate production in Ohio agroecosystems. Ph.D. diss. The Ohio State Univ., Columbus.
- Subler, S., C.M. Baranski, and C.A. Edwards. 1997. Earthworm additions increased short-term nitrogen availability and leaching in two grain-crop agroecosystems. Soil Biol. Biochem. 29:413–421.
- Subler, S., and A. Kirsch. 1998. Spring dynamics of soil carbon, nitrogen, and microbial activity in earthworm middens in a no-till cornfield. Biol. Fertil. Soils 26:243–249.
- Subler, S., R.W. Parmelee, and M.F. Allen. 1998. Earthworms and nitrogen mineralization in corn agroecosystems with different nutrient amendments. Appl. Soil Ecol. 9:299–306.
- Trojan, M.D., and D.R. Linden. 1998. Macroporosity and hydraulic properties of earthworm affected-soils as influenced by tillage and residue management. Soil Sci. Soc. Am. J. 62:1687–1692.
- Ward, A.D., J.L. Hatfield, J.A. Lamb, E.E. Alberts, T.J. Logan, and J.L. Anderson. 1994. The Management Systems Evaluation Areas Program: Tillage and water quality research. Soil Tillage Res. 30:49–74.
- Whalen, J.K., R.W. Parmelee, D.A. McCartney, and J.L. Vanarsdale. 1999. Movement of N from decomposing earthworm tissue to soil, microbial and plant N pools. Soil Biol. Biochem. 31:487–492.
- Wilkison, D.H., and D.W. Blevins. 1999. Observations on preferential flow and horizontal transport of nitrogen fertilizer in the unsaturated zone. J. Environ. Qual. 28:1568–1580.